2012

Performance Evaluation of Four Field-Scale Agricultural Drainage Denitrification Bioreactors in Iowa

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PERFORMANCE EVALUATION OF FOUR FIELD-SCALE AGRICULTURAL DRAINAGE DENITRIFICATION BIOREACTORS IN IOWA

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ABSTRACT. *Recently, interest in denitrification bioreactors to reduce the amount of nitrate in agricultural drainage has led to increased installations across the U.S. Midwest. Despite this recent attention, there are few peer-reviewed, field-scale comparative performance studies investigating the effectiveness of these denitrification bioreactors. The object of this work was to analyze nitrate removal performance from four existing bioreactors in Iowa, paying particular attention to potential performance-affecting factors including retention time, influent nitrate concentration, temperature, flow rate, age, length-to-width ratio, and cross-sectional shape. Based on a minimum of two years of water quality data from each of the four bioreactors, annual removal rates ranged from 0.38 to 7.76 g N m^{-3} bioreactor volume d^{-1}. Bioreactor and total (including bypass flow) nitrate-nitrogen load reductions ranged from 12% to 76% (mean 45%) and from 12% to 57% (mean 32%), respectively, removing from 0.5 to 15.5 kg N ha^{-1} drainage area. Multiple regression analyses showed that temperature and influent nitrate concentration were the most important factors affecting percent bioreactor nitrate load reduction and nitrate removal rate, respectively. This analysis also indicated that load reductions within the bioreactor were significantly impacted by retention time at three of the four reactors. More field-scale performance data from bioreactors of different designs and from multiple locations around the Midwest are necessary to further enhance understanding of nitrate removal in these systems and their potential to positively impact water quality.*

Keywords. *Denitrification bioreactor, Drainage, Nitrate, Water quality.*

Local water quality problems in the U.S. Midwest combined with national concerns about the hypoxic zone in the Gulf of Mexico require new approaches to improve agricultural drainage water quality (Turner and Rabalais, 1994; McMullen, 2001; IDNR, 2006; USEPA, 2007). Nitrate-nitrogen (NO_3^--N) loadings, one of the main contaminants of concern in agricultural drainage, can be reduced using a number of in-field and edge-of-field approaches, such as nutrient management, diversified crop rotations, and wetlands. However, in light of the U.S. Environmental Protection Agency's call for a 45% reduction in nitrogen (N) in the Mississippi River, a combination of approaches will be necessary (Dinnes et al., 2002; USEPA, 2007).

Denitrification bioreactors are a new remediation technology to reduce the amount of nitrate in agricultural drainage. The provision of additional carbon and maintenance of saturated conditions facilitates this "enhanced denitrification" process. In the U.S. Midwest, a handful of bioreactors have been installed in recent years, and lately, interest in these systems has grown, as evidenced by increased publicity in the mass media (e.g., Caspers-Simmet, 2010) and in the scientific literature (e.g., Schipper et al., 2010a; Strock et al., 2010; Woli et al., 2010).

Despite this interest in denitrification systems for NO_3^- removal, there are few peer-reviewed, field-scale performance studies investigating the effectiveness of agricultural drainage denitrification bioreactors. One of most comprehensive local studies is that of Jaynes et al. (2008), who showed that denitrification walls on the sides of a drainage tile pipe removed 55% of the NO_3^- load in the drainage when averaged over five years; however, the design of these walls was very different from most current drainage denitrification bioreactors (fig. 1). Other field-scale bioreactor work investigated hydraulic modeling (Chun et al., 2010) or provided performance data from early designs that lacked the control structure arrangement of more current bioreactors (Van Driel et al., 2006). Newer work by Woli et al. (2010) and Verma et al. (2010) in Illinois showed annual bioreactor load reductions of 23% to 98%. Based on this wide range, however, more continuous performance data from a number of denitrification bioreactors is needed for a more robust understanding of the potential contribution of these systems to water quality efforts.

Randall and Goss (2001) described controllable and uncontrollable factors for NO_3^- leaching from drainage, and there may be similar controllable and uncontrollable factors affecting bioreactor performance. A primary "controllable"

Submitted for review in February 2012 as manuscript number SW 9644; approved for publication by the Soil & Water Division of ASABE in September 2012.

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  ISSN 2151-0032

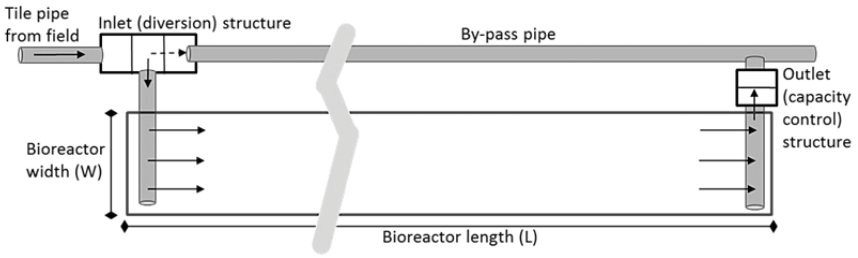

Figure 1. Generic top-view schematic of a denitrification bioreactor for agricultural drainage showing horizontal flow with distribution manifolds at bottom of bioreactor (not to scale; does not reflect the Pekin site studied here, as this site did not utilize control structures).

design factor is bioreactor length-to-width ratio (L:W), with most bioreactors in the Midwest tending to be long and narrow (i.e., high L:W). In one design model, the retention time was highly dependent on the length of the bioreactor, meaning that many resulting designs had L:W ratios of around 10 (length at least 30 m) (Christianson et al., 2011a). There has been little discussion in the literature about the effect of this ratio on performance. Another controllable design parameter is cross-sectional shape. Christianson et al. (2010a) found there was no difference in NO_3^- removal between a trapezoidal cross-section versus a rectangular cross-section in pilot-scale denitrification bioreactor experiments. Although several trapezoidal cross-section denitrification systems have been installed (Christianson et al., 2009; Schipper et al., 2010b), the specific design effect of cross-sectional shape has not been investigated at the field scale.

Retention time, or the relationship between the media porosity, active flow volume, and flow rate through the reactor, is a performance parameter that combines controllable design factors with uncontrollable environmental elements. The selection of fill media and the design dimensions of the bioreactor are controllable, but the variable flow rate and, to some extent, the depth of water in the reactor make designing for a specific retention time challenging (Christianson et al., 2011a; Woli et al., 2010). The USDA-NRCS interim design standard for denitrifying bioreactors in Iowa specifies a retention time that allows sufficient reduction in NO_3^- concentration (USDA-NRCS, 2009); however, an "adequate" retention time may most likely vary based on hydraulic loading and temperature (Christianson et al., 2011a).

Significant environmental factors that may affect bioreactor NO_3^- removal performance include influent NO_3^- concentration, hydraulics, temperature, and bioreactor age. There has been discussion in the literature regarding the impact of influent NO_3^- concentration on removal. Some reports have indicated that NO_3^- removal rates will be constant regardless of influent NO_3^- concentration (zero-order reaction) (Gibert et al., 2008; Robertson, 2010), while others have found that increased NO_3^- concentrations increase the removal rate (first-order reaction) (Chun et al., 2009). Robertson (2010) noted that NO_3^- removal followed zero-order kinetics due to insensitivity to influent NO_3^- concentrations; the reaction may be controlled by an independent parameter, such as labile carbon availability. This carbon availability can be impacted by bioreactor hydraulics, with Woli et al. (2010) noting that several dry periods in a bioreactor may have precipitated greater labile carbon availability and thus high removal for subsequent high flow events. *In situ* temperatures can also be important, with NO_3^- removal typically increasing by a factor of approximately 2 for every 10°C increase in temperature (i.e., $Q_{10} \approx 2$) (Cameron and Schipper, 2010; Warneke et al., 2011). Finally, longevity can impact performance, but there seem to be no field-scale bioreactors in operation long enough to have failed due to carbon exhaustion (Schipper et al., 2010a). Moorman et al. (2010) and Long et al. (2011) documented denitrification systems that had sufficient carbon to continue operation after nine and fourteen years, respectively.

Although a number of bioreactors are now in operation in the U.S. Midwest, there is a lack of comprehensive peer-reviewed performance evaluations from multiple sites. Past work highlighted the potential for promising NO_3^- removal from these systems, but performance optimization and prediction require more advanced analysis techniques. The objective of this work was to analyze NO_3^- removal performance from four existing bioreactors in Iowa with particular attention paid to the factors affecting performance (i.e., retention time, L:W, cross-section, influent concentration, temperature, age, and flow rate). Multiple regression analysis was then used to identify which of these factors most affected NO_3^- removal performance. It was hypothesized that denitrification bioreactor design parameters and field conditions affect *in situ* performance. Lastly, a cost analysis was included to allow economic comparison of bioreactors with other water quality technologies.

METHODS

Four woodchip-based denitrification bioreactors in Iowa, each with a different design and drainage treatment area, were used for this comparison (table 1). The Greene County and Hamilton County bioreactors were monitored by the Iowa Soybean Association Environmental Programs and Services (ISA), while the Northeast Research and Demonstration Farm (NERF) and Pekin bioreactors were monitored by Iowa State University researchers. The parameters under analysis included length-to-width ratio (L:W), cross-sectional shape, flow rate, temperature, age, and NO_3^--N influent and effluent concentration, with the derivative factors of retention time and NO_3^- removal rate calculated. Contributing areas (i.e., drainage treatment areas) were determined based on knowledge of the existing

Table 1. Description of four agricultural drainage denitrification bioreactors in Iowa used in investigation.

Bioreactor	Location	Date of Installation	Drainage Treatment Area (ha)	Length (m)	Width (m)	Depth (m)	Volume (m^3)
Pekin	Southeast Iowa	August 2002	1.3	30	0.5	1.2	18
NERF	Northeast Iowa	April 2009	14.2	36.6	4.6 top, 2.4 bottom	1.0	128
Greene Co.	Central Iowa	August 2008	19.0	15.2	7.6	1.1	127
Hamilton Co.	Central Iowa	June 2009	20.2	30.5	3.7	0.9	102

tile drainage network; however, when this was unknown (e.g., NERF), the contributing area was assessed based on estimated annual subsurface water transmission. Although the drainage area for only the Pekin bioreactor was exactly known (i.e., a small-scale research plot), parameters such as kg N lost ha^{-1} and water drainage depths were nevertheless calculated to allow comparisons between this and other field-scale studies.

PEKIN, IOWA

As one of the oldest denitrification bioreactors in the state, the bioreactor in Pekin, Iowa, yielded this study's longest data record. Installed in August 2002, it was filled with a mixture of gravel and woodchips (approximately 60% woodchips by volume) and received drainage from a research plot, hence the small treatment area of 1.3 ha (plots of 91 × 140 m). Based on visual observation, the woodchips and gravel rocks were comparable in size; most pieces were approximately 25 to 50 mm mean particle size. In addition to the gravel used in the fill, this reactor differed from the other three bioreactors in that it only had one control structure on the inlet side rather than two structures and did not have a bypass line. Drainage from the research plot was routed through the inlet structure into a sump from where it was pumped into the bioreactor (i.e., the single inlet control structure was not used to divert flow, as at the other sites). Bioreactor outflow free-flowed into an outlet sump, where it was pumped through a flowmeter (Neptune T-10 meters for both inflow and outflow). The bioreactor likely received lateral flow from neighboring research plots, although inflow and outflow were generally within 20% of each other for six of the seven years under investigation. Flow-proportional samples were collected from both sumps from late spring through late summer from 2005 to 2011 with sampling procedures described by Lawlor et al. (2008). For each research plot at the site, drainage was routed to a subsurface vertical culvert, where it was pumped through a flowmeter; backpressure due to this meter forced a small fraction (approximately 0.25%) through plastic tubing into a glass collection bottle (Lawlor et al., 2008). Samples were frozen until they were transported in a cooler to the laboratory. Additionally, from 2009 onward, the sample bottles were acidified with dilute sulfuric acid prior to sample collection. Nitrate-N analysis for this site was done using second-derivative spectroscopy in the Wetland Research Laboratory at Iowa State University (Crumpton et al., 1992). Although samples from the other bioreactors were analyzed in different laboratories with other methods (e.g., cadmium reduction, ion chromatography), these methods compare well, with reported agreements of r^2 = 0.99 (Crumpton et al., 1992; Ferree and Shannon, 2001).

NERF (NORTHEAST RESEARCH AND DEMONSTRATION FARM)

Located in northeast Iowa, the 100% woodchip NERF bioreactor was installed in April 2009 with a trapezoidal cross-section. The NERF has been the location of a number of agricultural field studies since 1976. The 14.2 ha NERF bioreactor drainage area was in a corn and soybean rotation during the investigation period. The estimated drainage area was based on visual observation of the site in combination with the percentage of precipitation occurring as total measured flow in the tile main. The woodchips used at this installation were obtained from a local supplier and were similar in size and shape to the woodchips described by Christianson et al. (2010b) (i.e., greater than 50% by mass fall between the 9.5 to 25 mm particle size, carbon-to-nitrogen ratio similar to 250). After installation, all drainage flow was routed to bypass the reactor until flow monitoring equipment was installed in October 2009. Bypass flow depth in the inflow structure and bioreactor flow depth in the outflow structure were continuously logged with pressure transducers (WL16 Water Level Loggers, Global Water Instrumentation, Inc., from October 2009 to April 2011; Levelogger Junior, Solinst, from April 2011 to August 2011). Outflow control structure transducer data were used for both bioreactor inflow and outflow values by assuming bioreactor inflow equaled bioreactor outflow. Transducer depth data were reduced to daily average values to increase data workability and to allow synchronization with sample event collection days. These daily transducer depths were adjusted based on stop log height in the structures to give flow depth over the stop logs. During periods of pressure transducer logging failure, depths logged by area velocity meters (2150 area velocity module, Teledyne ISCO) installed upstream of both structures were used. Flow equations developed by Chun and Cooke (2008) (eq. 1) for 15 cm control structures were used to convert flow depths to flow rates for data until 8 April 2011, when 45° V-notch weirs were installed in the structures and a corresponding V-notch weir flow equation was used (eq. 2):

$$\begin{aligned} Q &= 0.02\left(L - 0.437H\right)H^{1.48} \quad \text{for } H \le 0.44L \\ Q &= 0.027LH^{1.2} \quad \text{for } H > 0.44L \end{aligned} \tag{1}$$

$$Q_{v1} = 415.4\left(h_w + 0.0519\right)^{2.5} \tag{2}$$

where Q is the flow rate in the structure (L s^{-1}), L is the stop log width (cm), H is the flow depth above the stop log (cm), Q_{v1} is the discharge over the weir (gpm), and h_w is the head over the weir (ft). The coefficients in equation 2 were developed through calibration of the V-notch weir (K. Heikens, personal communication, 2011).

The bypass stop logs in the inflow structure and the capacity control stop logs in the outflow structure were periodically managed during the study. In flow calculations, the total allowable flow into the system was capped based on the drainage pipe size and estimated tile slope in the field. Conservation of NO_3^--N mass in the bypass line was assumed for this and the following two reactors. The total inflow and outflow loads consisted of the inflow bioreactor load plus the bypass load and the outflow bioreactor load plus the bypass load, respectively.

Grab samples from the control structures were collected by the farm staff at least twice weekly, stored at 4°C, and analyzed in the Iowa State University Agricultural and Biosystems Engineering Water Quality Research Laboratory (ISU ABE WQRL) for NO_3^--N + NO_2^--N using a cadmium-reduction method (Quick-Chem 8000 automated analyzer, Lachat Instruments). Additionally, sulfate samples were analyzed in the ISU ABE WQRL using Hach test method 8051 (USEPA SulfaVer 4 method; barium sulfate precipitation). Water temperature of the samples was recorded immediately after sample collection from the structures with a handheld digital thermometer (Fisher Scientific).

GREENE COUNTY, IOWA

The Greene County bioreactor was installed in summer 2008 in central Iowa with the lowest L:W in this comparison. The 19 ha drainage treatment area was continuously cropped in a corn and soybean rotation, and the bioreactor was fed by a 30 cm tile pipe. The contributing drainage area was estimated from a GPS-based topographic survey plus landowner input. The Greene County bioreactor was designed to provide 4 h of retention time while treating 20% of the expected peak flow rate (Christianson et al., 2011a). The expected peak flow rate was calculated based on a 20 cm tile diameter, as the installed 30 cm tile pipe was thought to be oversized to compensate for a surface intake. The woody media used here consisted of a mix of shredded material and chips, both of which were described in more detail by Christianson et al. (2010b). Logging pressure transducers (American Sensor Technologies) in the inflow and outflow structures were used to determine bypass and bioreactor flow, respectively. On selected sampling dates, a 19 L (5 gal) bucket and stopwatch were used to verify outflow rate, and the depth of water in the structures was manually checked. In flow calculations, these manual bucket and depth measurements were used as calibration points; where transducer data were missing, the manual water depth measurements were used with a linear interpolation to estimate flows. For example, the inflow transducer stopped working in March 2010 and was not replaced until January 2011; thus, these data were interpolated.

A 45° V-notch weir was installed in the structures on 31 March 2010; equations from Chun and Cooke (2008) for 30 cm control structures (eq. 3) were used until this date, while equation 4 was used to calculate flow when the V-notch weirs were in place:

$$\begin{aligned} Q &= 0.02\left(L - 0.74H\right)H^{1.48} \quad \text{for } H \le 0.27L \\ Q &= 0.021LH^{1.37} \quad \text{for } H > 0.27L \end{aligned} \tag{3}$$

$$Q_{v2} = 4.28C_e \tan\left(\frac{\theta}{2}\right)\left(h_1 + k_h\right)^{2.5} \tag{4}$$

where the terms in equation 3 are as described for equation 1, and Q_{v2} is the discharge over the weir ($ft^3\ s^{-1}$), C_e is the effective discharge coefficient, θ is the V-notch angle (degrees), h_1 is the head over the weir (ft), and k_h is a head correction factor. For a 45° V-notch weir, C_e and k_h are approximately 0.58 and 0.005, respectively, with the original equation (eq. 4) based in U.S. customary units (USBR, 2001). A compound weir equation was used for several dates in June and August 2010 when the flow depth was greater than the depth of the V. This calculation allowed flow calculation for the full V height (16 cm), with the additional flow calculated by equation 3 for the marginal depth above this V height.

Grab samples were transported to the laboratory in a cooler on ice and were analyzed either the day of collection or were refrigerated at 4°C until analysis within 48 h. In addition to analysis for NO_3^--N, the Greene County and Hamilton County bioreactor grab samples were analyzed for dissolved oxygen (DO) and sulfate at the Des Moines Water Works or at the ISA's internal laboratory beginning April 2011 (ion selective probe method, Standard Method 4500-NO3D; or ion chromatography, EPA Method 300.0).

HAMILTON COUNTY, IOWA

The Hamilton County bioreactor was installed in central Iowa in 2009 with surface dimensions similar to the NERF bioreactor. However, the Hamilton County reactor had a rectangular cross-section and received drainage from a larger area than the NERF site. The Hamilton County bioreactor was designed to have a 4 h retention time while treating 20% of the expected peak flow from a 15 cm tile (Christianson et al., 2011a). The cropping rotation was soybean-corn-corn, and the contributing drainage area was estimated based on aerial photos of the tile installation in conjunction with 30 m resolution topographic maps and landowner input. Woodchip fill at this reactor was similar to the chips at the NERF site (e.g., the majority of chips were 9.5 to 25 mm particle size based on visual observation) although from a different local supplier. Bioreactor flow monitoring, calculations, and sample analyses were similar to the Greene County site except with 15 cm structures used rather than 30 cm structures. The Chun and Cooke (2008) flow equations (eq. 1) were used for the pressure transducer data until 19 August 2010, when 45° V-notch weirs were installed (eq. 4). After removing several periods of bioreactor flooding in 2010 from the dataset, there was no need for compound weir calculations.

MULTIPLE REGRESSION ANALYSIS

Multiple regression analysis (SAS Proc Reg; SAS Institute, Inc., Cary, N.C.) of the four bioreactors was undertaken to determine which measured environmental parameters were key drivers of nitrate removal. A regression model describing the percentage load reduction and a regression model describing the removal rate were developed for each site. Explanatory factors in both models included retention

time, influent NO_3^--N concentration, influent water temperature, flow rate, and bioreactor age. Retention time was calculated as the active flow volume (i.e., geometric volume including both chips and voids) multiplied by an assumed porosity of 0.6 for all sites (Ima and Mann, 2007) divided by the reactor flow rate. The active flow volume was based on the flow depth, assuming a linear head difference between the water depths in the inflow and outflow structures. The reactor flow rate was the incremental difference in outflow volume between two sampling events divided by the change in time between the events. Because nitrate samples were not collected every day, daily incremental flow volumes occurring after the previous sample event and including the day of the sample event of interest were summed; this cumulative flow volume was used with the latter sample concentration for the mass NO_3^--N calculation at that latter date. Despite not collecting samples daily, it was thought that the at least monthly sampling scheme (and often at least several samples per month) during active flow periods still provided reasonable accuracy (i.e., greater than 70% probability of being within ±15% of the "true" annual load, following Wang et al., 2003). Drainage area-based loads (kg N ha^{-1}) were calculated by dividing the mass load from each sample date by the drainage treatment area. Annual average daily removal rates (g N m^{-3} d^{-1}) were calculated from the annual summed mass of NO_3^--N removed divided by the entire bioreactor volume and the difference between the first and last sample date for each year. Annual percentage load reduction was calculated by dividing the difference of the annual inflow and outflow loads by the annual inflow load. Likewise, annual flow-weighted percent concentration reduction was calculated by dividing the difference of the annual flow-weighted influent and effluent concentrations by the annual flow-weighted influent concentration. In the regression analyses, sample event-based values rather than annual values were used as observations. In other words, the dependent variables in the regressions (percent load reduction and removal rate) were based on sample event influent/effluent loads and the change in time between two given sample events.

Two regression analyses (percent load reduction and removal rate) were additionally performed for a combined dataset from all four reactors. Data for these regressions were pooled based on the number of observations at each site, meaning that the NERF bioreactor, with the most sample events, had the heaviest weight. Observations with missing values (e.g., temperature was not recorded at all sample events) were not included, resulting in populations of $n = 142$ for these combined regressions. These comprehensive models included the above explanatory factors as well as the L:W and a factor for cross-sectional shape. Regression procedure results included parameter estimates and associated standard error for each of these independent factors, along with an indication of model fit (R^2). The significance of each independent parameter in the site-specific models was determined at $\alpha = 0.01$, 0.05, and 0.10. In the combined dataset models, a stepwise selection procedure was used to eliminate parameters from the model unless they were significant at the $\alpha = 0.05$ statistical level. Stepwise regression is a modification of a forward selection technique in which significant variables are added to the model one at a time, although they may not remain as more variables are added.

RESULTS AND DISCUSSION

NITRATE REMOVAL

The annual influent flow-weighted NO_3^--N concentrations were generally lowest in the Pekin bioreactor (annual means of 1.23 to 8.54 mg NO_3^--N L^{-1}), with this bioreactor also having the three lowest flow-weighted effluent concentrations (0.63, 1.31, and 1.89 mg NO_3^--N L^{-1}) (table 2). The annual flow-weighted influent concentrations were fairly comparable at the other three sites, which ranged from 7.70 to 15.18 mg NO_3^--N L^{-1} (table 2). Influent values usually peaked in summer months at greater than 15 mg NO_3^--N L^{-1} (figs. 2b, 2c, and 2d, non-flow-weighted concentrations). Nevertheless, effluent concentrations at the Hamilton Co. and Greene Co. sites were less than 10 mg NO_3^--N L^{-1} for all but one sample at each site (13 May 2010 and 27 June 2011, respectively). Effluent concentrations from the NERF bioreactor exceeded this 10 mg NO_3^--N L^{-1} maximum contaminant level (USEPA, 2011) more frequently, which was also reflected in the elevated NERF annual flow-weighted effluent concentrations (8.51 and 11.62 mg NO_3^--N L^{-1}) compared to the other sites.

Annual bioreactor NO_3^- removal rates ranged from 0.38 to 7.76 g N m^{-3} d^{-1} (table 2). These values were similar to the range of published literature, with a review by Schipper et al. (2010a) reporting NO_3^- removal rates of 2 to 22 g N m^{-3} d^{-1} for a variety of denitrification bed systems. More specific to drainage treatment, Christianson et al. (2011b) reported rates of 3.8 to 5.6 g N m^{-3} d^{-1} in pilot-scale work, and Woli et al. (2010) reported a rate of 6.4 g N m^{-3} d^{-1} for a field-scale bioreactor in Illinois.

Bioreactor influent loads ranged from 0.8 to 34.7 kg N ha^{-1}, while effluent loads were between 0.2 and 29.7 N ha^{-1} for all four bioreactors in all years (table 2, fig. 2). When the bypass flow volume at the sites was considered in addition to the bioreactor flow volume (i.e., "total" loads as opposed to "bioreactor" loads), the resulting total inflow and outflow loads were 1.2 to 50.1 N ha^{-1} and 0.6 to 34.6 kg N ha^{-1}, respectively (table 2). The Hamilton Co. bioreactor had the highest total percent load reductions at greater than 48% in both years, although this only equated to 8.1 and 0.6 kg N ha^{-1} removed. The Greene Co. reactor removed the greatest annual load with removal of 15.5 kg N ha^{-1} in 2009; however, this was only a 30.9% total load reduction.

Because the Pekin bioreactor inflows and outflows were pumped through flowmeters and there was no bypass flow, these inflow and outflow depths, rather than bioreactor and total depths, are shown in table 2; lateral seepage at the site likely accounts for the discrepancy between these values in the percent flow treated column. Neglecting this site, the NERF bioreactor treated the highest percentages of water (greater than 90%), although this site had the lowest percentage bioreactor and total load reductions (table 2). The Hamilton Co. bioreactor also treated the majority of drain-

Table 2. Annual influent and effluent NO_3^--N concentrations and loads by bioreactor or total (including bypass) and NO_3^--N removal rates for four denitrification bioreactors in Iowa.

Site and Water Year[a]	Nitrate-N Concentration[b]			Nitrate-N Load							Water Depth		
	In (mg NO_3^--N L^{-1})	Out (mg NO_3^--N L^{-1})	Reduction (%)[c]	Bioreactor In (kg N ha^{-1} drainage area)	Bioreactor Out (kg N ha^{-1} drainage area)	Mean Bioreactor Reduction (%)[c]	Removal Rate (g N m^{-3} d^{-1})[d]	Total In (kg N ha^{-1} drainage area)	Total Out (kg N ha^{-1} drainage area)	Mean Total Reduction (%)[c]	Bioreactor[e] (cm)	Total[f] (cm)	Depth Treated (%)
Pekin													
2004	4.21	1.89	55.2	5.0	2.8	43.7	1.07				11.9	14.6	81.7
2005	4.98	2.22	55.5	1.2	0.7	43.5	0.75				2.3	2.6	88.6
2006	8.54	4.66	45.4	33.6	21.1	37.4	3.78		NA[g]		39.4	49.5	79.5
2007	2.86	2.46	14.0	14.8	8.4	43.8	2.53				51.9	51.6	101
2008	3.84	2.49	35.2	7.1	5.0	29.1	0.57				18.5	20.6	89.8
2009	1.23	0.63	49.1	7.4	5.8	22.0	0.67				60.5	56.2	108
2010	1.88	1.31	30.5	2.0	0.5	74.0	0.38				10.5	2.3	456
NERF													
2009	9.93	8.51	14.3	34.7	29.7	14.6	1.56	37.3	32.2	13.6	34.0	37.4	90.9
2010[h]	13.18	11.62	11.9	21.4	18.9	11.7	0.86	21.9	19.4	11.5	18.1	18.4	98.6
Greene Co.													
2008[i]	15.18	4.97	67.2	20.2	6.5	68.0	7.76	41.4	27.6	33.3	20.9	39.2	53.4
2009	7.70	4.67	39.4	33.6	18.1	46.0	6.69	50.1	34.6	30.9	44.6	65.2	68.4
2010[j]	9.55	6.18	35.2	1.6	0.8	50.4	0.41	2.9	2.1	27.3	1.5	3.0	50.8
Hamilton Co.													
2009	7.74	1.92	75.2	10.8	2.6	75.7	5.02	14.4	6.3	56.6	16.2	18.7	87.0
2010[k]	9.59	2.47	74.3	0.8	0.2	73.9	0.42	1.2	0.6	48.6	0.9	1.2	73.2

[a] Water year is defined as the year beginning on 1 October and ending 30 September the following year.
[b] Annual flow-weighted concentrations.
[c] Reduction (%) is calculated as the reduction between annual flow-weighted inflow/outflow concentrations or annual loads, not the mean of reductions of individual sample events.
[d] Removal rate is based on the annual summed mass of NO_3-N removed divided by the entire bioreactor volume and the difference between the first and last sample dates for each year.
[e] For the Pekin bioreactor, this was bioreactor inflow depth.
[f] For the Pekin bioreactor, this was bioreactor outflow depth.
[g] Not applicable because the Pekin bioreactor had no bypass.
[h] Through 22 August 2011.
[i] No flow monitoring until 1 January 2009.
[j] Through 25 July 2011.
[k] Through 6 July 2011.

age water in both years of performance (greater than 73% treated) but was able to treat these waters with higher load reductions than at the NERF site. This difference between the reactors indicates that, while it is useful to route as much drainage as possible through the bioreactor, there may be a useful compromise between treating a slightly lower volume of water at a better treatment rate. The impact of treating too little of the drainage can been seen with data from the Greene Co. reactor. This reactor treated between 50.8% and 68.4% of drainage in three years of operation, which greatly reduced its treatment efficiency from 46% to 68% (bioreactor load reduction) to less than 34% total load reduction. Note that rainfall in the 2010 water year was lower than the long-term averages at the Greene Co. and Hamilton Co. sites, which may account for the low amount of drainage in those site-years.

In addition to the percentage of drainage water treated, reactor hydraulics were another important consideration for bioreactor performance. Based on preliminary tracer testing at the NERF site, flow short-circuiting within the reactor was very likely (L. Christianson, unpublished data). Short-circuiting causes a portion of the drainage to remain in the bioreactor for a shorter period than indicated by the theoretical retention time, thus decreasing the reactor's NO_3^- removal potential. This furthermore accounts for the poor performance of the NERF bioreactor.

Additional Parameters

Bioreactor water temperature for all sites peaked in late summer months at typically greater than 15°C and was at its lowest around March of each year at less than 3°C (fig. 3). The annual flow-weighted inflow temperature ranged from 6.08°C to 8.69°C (mean 7.09°C) for these three sites in these years. Like temperature, bioreactor influent DO fluctuated annually, with the highest influent DO in early spring months (≥8.5 mg DO L^{-1}) and the lowest in summer (mid-July through late August, typically <5 mg DO L^{-1}) (fig. 3). Regardless of influent DO concentration, this parameter was always reduced to less than 2.4 mg DO L^{-1} (and usually much less), indicating that conditions conducive to denitrification were present within these bioreactors.

Sulfate reduction was also documented in the Hamilton Co., Greene Co., and NERF bioreactors, although not continuously during all sample events (fig. 4). Sulfate reduction is due to an excess of reducing capacity in the reactors once the influent NO_3^- is removed (Blowes et al., 1994). This process was most notable in winter months in the Hamilton Co. and Greene Co. reactors (November 2009 and December 2008, respectively) when influent NO_3^- was reduced to nearly zero from concentrations of approximately 8 mg NO_3^--N L^{-1} and greater than 11 mg NO_3^--N L^{-1} at the two sites, respectively. More continuous sulfate reduction was documented in the NERF bioreactor from late August to mid-October 2010 when influent NO_3^- was similarly

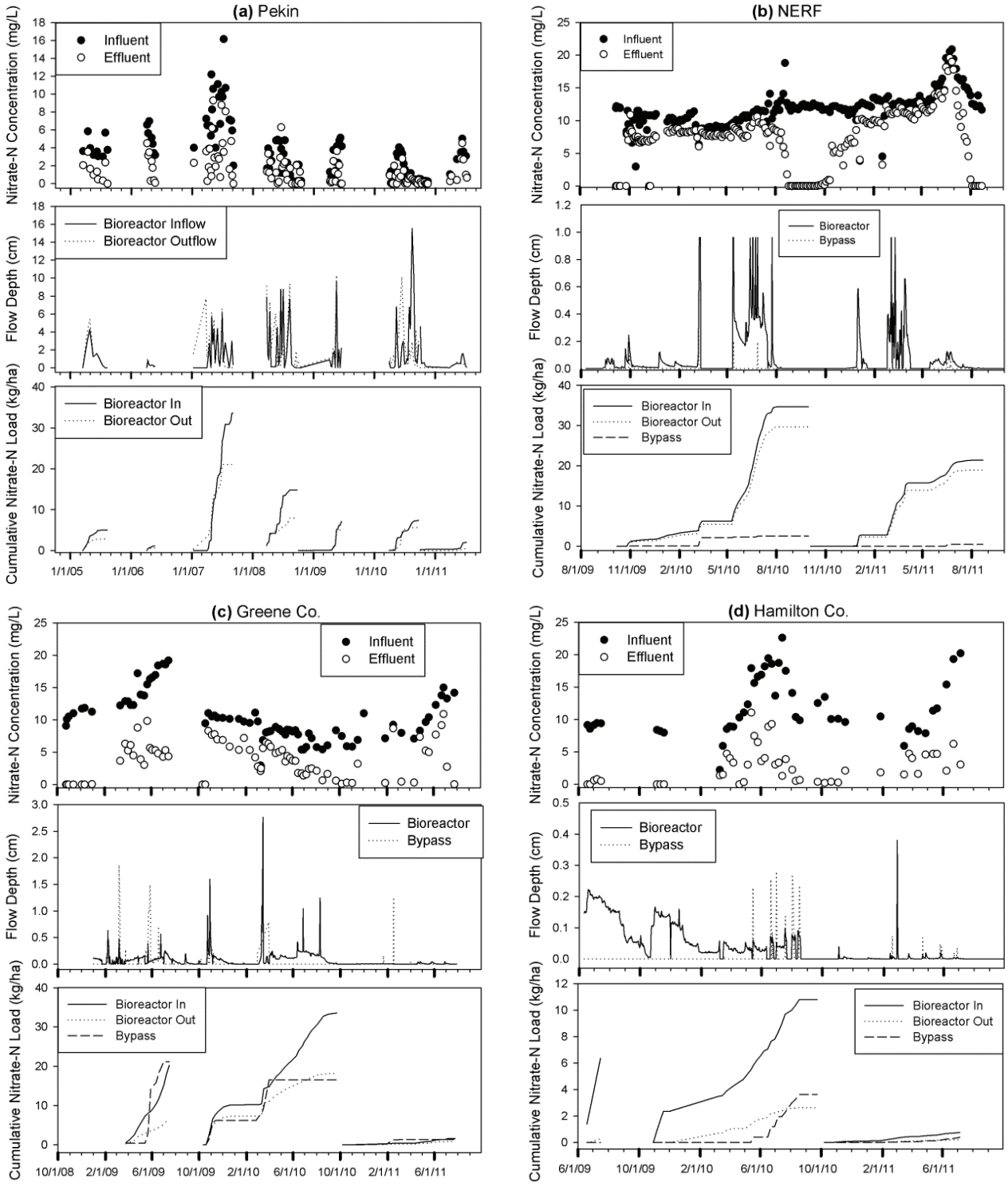

Figure 2. Influent and effluent NO_3^--N concentrations, bioreactor and bypass flows, and cumulative NO_3^--N loads for four denitrification bioreactors in Iowa; flow depths are normalized by drainage treatment area (i.e., they are not the depths over weir). Note different scales on *y*-axes.

reduced. The low flow rate through this reactor at this time (fig. 1b) was indicative of high retention times and thus complete NO_3^- reduction and subsequent sulfate reduction.

MULTIPLE REGRESSION RESULTS

Individual regression models for the four bioreactors revealed that the dependent parameter percent bioreactor load reduction was most strongly positively correlated with temperature (correlation at $\alpha = 0.01$ significance for two of three reactors where this parameter was measured; table 3). Retention time was also noticeably correlated, although this relationship was strongly significant at only one site (NERF at $\alpha = 0.01$ level; table 3). Bioreactor age had a significant impact on percent load reduction for three of the four bio-

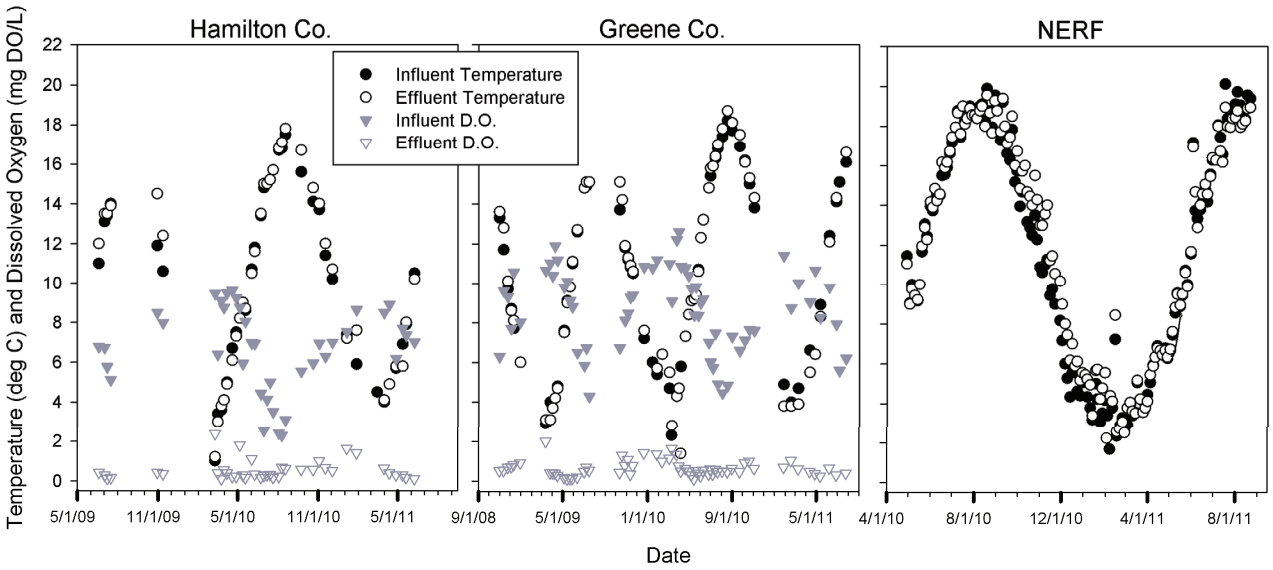

Figure 3. Influent and effluent water temperature from three bioreactors and influent and effluent DO from two bioreactors in Iowa.

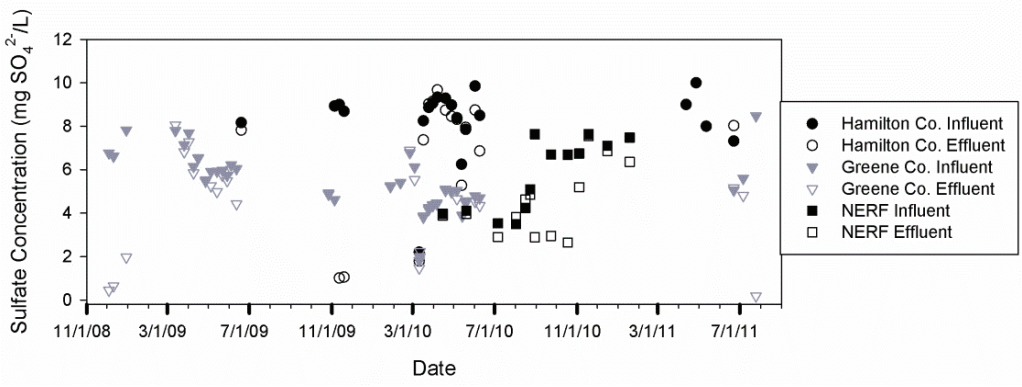

Figure 4. Influent and effluent sulfate concentrations for three denitrification bioreactors in Iowa.

Table 3. Percent N load reduction regression model parameter estimates (standard errors in parentheses) for independent factors of retention time, influent NO_3- concentration, temperature, flow rate, and age at four denitrification bioreactors in Iowa.[a]

Site (No. of Observations)	Model Intercept	Retention Time (h)	Influent Concentration (mg NO_3^--N L^{-1})	Temperature (°C)	Flow Rate (m^3 h^{-1})	Age (months)	R^2
Pekin	26.88	0.39***	0.44	NA[b]	-5.04**	0.41**	0.34
(*n* = 48)	(18.40)	(0.23)	(1.32)		(2.40)	(0.18)	
NERF	17.53	0.73*	-4.68*	1.81*	-0.31	2.02*	0.74
(*n* = 76)	(11.30)	(0.13)	(1.11)	(0.34)	(0.20)	(0.57)	
Greene Co.	50.68*	0.74**	-1.47	4.74*	-0.57	-1.78**	0.49
(*n* = 43)	(18.16)	(0.34)	(1.05)	(0.87)	(0.44)	(0.70)	
Hamilton Co.	77.58*	0.04	-0.49	2.26***	-0.52	-1.40	0.36
(*n* = 23)	(20.77)	(0.26)	(1.26)	(1.22)	(1.48)	(1.69)	

[a] Asterisk (*) indicates significance at $\alpha = 0.01$, ** indicates significance at $\alpha = 0.05$, and *** indicates significance at $\alpha = 0.10$.
[b] Temperature was not measured at the Pekin bioreactor.

reactors; this factor did not have a consistently positive or negative effect, which confounded its importance (table 3). Flow rate and influent NO_3^- concentration were only significant at the Pekin and NERF sites, respectively (table 3).

The NO_3^- removal rate regression models showed that the removal rate metric in terms of g N m^{-3} d^{-1} was most significantly affected by flow rate and influent NO_3^- concentration (table 4). Flow rates were strongly significant at the Green Co., Hamilton Co., and NERF sites ($\alpha = 0.01$), while the influent concentration had the same level of significance only at the Green Co. and Hamilton Co. reactors (table 4). Importantly, this dependence of removal rate on flow rate was likely an artifact of calculation, as similar original raw data were required in the computation of both parameters. The positive correlation of removal rate with influent concentration at a minimum $\alpha = 0.05$ level for three of the bioreactors may help clarify the reaction kinetics. The parameter estimates for these three reactors indicated that a 1 mg NO_3^--N L^{-1} increase in influent concentration increased the removal rate by 0.44 to 1.25 g N m^{-3} d^{-1},

Table 4. N removal rate regression model parameter estimates (standard errors in parentheses) for independent factors of retention time, influent NO_3- concentration, temperature, flow rate, and age at four denitrification bioreactors in Iowa.[a]

Site (No. of Observations)	Model Intercept	Retention Time (h)	Influent Concentration (mg NO_3^--N L^{-1})	Temperature (°C)	Flow Rate (m^3 h^{-1})	Age (months)	R^2
Pekin	-6.67	0.03	1.02**	NA[b]	1.33***	0.07	0.21
(*n* = 48)	(6.01)	(0.07)	(0.43)		(0.78)	(0.06)	
NERF	1.35	-0.02	-0.11	0.08**	0.23*	0.00	0.76
(*n* = 76)	(1.28)	(0.01)	(0.13)	(0.04)	(0.02)	(0.06)	
Greene Co.	-14.70*	0.05	1.25*	0.57**	0.77*	-0.11	0.77
(*n* = 43)	(5.33)	(0.10)	(0.31)	(0.25)	(0.13)	(0.21)	
Hamilton Co.	-5.66**	0.02	0.44*	0.21	1.99*	-0.09	0.97
n = 23)	(2.30)	(0.03)	(0.14)	(0.14)	(0.16)	(0.19)	

[a] Asterisk (*) indicates significance at $\alpha = 0.01$, ** indicates significance at $\alpha = 0.05$, and *** indicates significance at $\alpha = 0.10$.
[b] Temperature was not measured at the Pekin bioreactor.

Table 5. Percent N load reduction and N removal rate regression model parameter estimates (standard errors in parentheses) for the pooled dataset from four denitrification bioreactors in Iowa (*n* = 142).

	Model Intercept	Retention Time (h)	Influent Concentration (mg NO_3^--N L^{-1})	Temperature (°C)	Length-to-Width Ratio	Cross-Sectional Shape	Flow Rate (m^3 h^{-1})	Age (months)	R^2
Percent N	17.98	0.48	-[a]	2.20	3.39	-75.77	-[a]	-[a]	0.59
load reduction	(5.36)	(0.09)		(0.32)	(0.81)	(9.64)			
N removal rate	1.96[b]	-[a]	0.54	0.34	-[a]	-8.44	0.44	-0.30	0.63
	(2.27)		(0.13)	(0.08)		(0.98)	(0.05)	(0.07)	

[a] Parameter was not significant at the $\alpha = 0.05$ and thus was not included in the model.
[b] Intercept was not significant at the 0.05 level, although it was nevertheless included in the model as the linear regression intercept.

assuming other parameters were held constant. This nearly proportional 1:1 relationship (on average 1:0.9) and its significance point strongly to first-order kinetics for these data where reaction rate is dependent on the availability of the reactant (Tchobanoglous et al., 2003). A zero-order relationship would likely not have shown significance between these variables. Importantly, however, this regression approach assumed that all explanatory parameters other than influent concentration were held constant, which was a major limitation of this approach. Under field conditions, reaction kinetics may be masked or convoluted by a number of environmental factors. Schipper et al. (2010a) noted that denitrification systems may functionally use zero-order kinetics, although first-order reactions most closely described a drainage bioreactor in Illinois (Chun et al., 2010) and an enhanced-denitrification wetland system in California (Leverenz et al., 2010).

Removal rates generally increased with increasing temperature, and this influent water temperature was a significant performance factor at the NERF and Greene Co. bioreactors ($\alpha = 0.05$) (table 4). The effect of temperature on a reaction may be express as Q_{10}, the factor by which the removal rate increases for every 10°C increase in temperature. Here, the removal rate model estimates for temperature indicated that the Q_{10} for these reactors ranged from 0.8 to 5.7. This range overlaps past work in this field showing Q_{10} values of approximately 0.8 to 2.4 (Cameron and Schipper, 2010; Warneke et al., 2011), with the higher value here more similar to extrapolations from work by Robertson and Merkley (2009) and Van Driel et al. (2006), which showed Q_{10} values from 2 to 3 with an extrapolation from Robertson et al. (2008) yielding a Q_{10} of 5.0.

The use of the stepwise selection procedure in the development of the combined dataset models (i.e., all bioreactors analyzed together) allowed introduction of an explanatory parameter to the model only if it was significant at the $\alpha = 0.05$ level (table 5). These regression models yielded R^2 values of 0.59 and 0.63 for the percent load reduction and the removal rate model, respectively. Not surprisingly based on the individual bioreactor models, temperature was significant in both combined dataset models. The significance of the length-to-width ratio and cross-section parameters in the percent load reduction model indicated that there was a significant difference between the four sites, a difference that could also be seen by comparing the annual bioreactor reductions in table 2. Testing of these parameters in this pooled dataset was intended to elucidate performance differences due to design factors, but any such potential differences between designs may have been complicated by the significance of the environmental factors.

Regression diagnostics were performed for all the models to detect possible multicollinearity (i.e., close relationships between explanatory variables) and correlated errors associated with the time-series nature of these data. In testing for multicollinearity, a variance inflation factor of greater than 10 generally indicates that two or more variables are linear combinations of each other. Here, the variance inflation factors were less than 10 for all variables in all regression models and were generally less than 4. Durbin-Watson tests (at 5% significance) were used to detect correlated errors between temporally adjacent observations. These tests showed there was a time dependency for these models, as values closer in time may have been related. Such autocorrelated errors are a limitation of this regression modeling approach for these data.

COST

Because comprehensive cost data were not available for the NERF and Pekin bioreactors, cost data from the Greene Co. and Hamilton Co. bioreactors along with four additional bioreactors in Iowa were collected to evaluate a full range of costs. The total installation cost for six bioreactors in Iowa ranged from $4,390 to $11,820 and from $194.72 ha^{-1} to $585.64 ha^{-1} (table 6). Treated areas ranged

Table 6. Installation costs for six denitrification bioreactors treating agricultural drainage in Iowa with each major cost component and cost per drainage treatment area shown.

Reactor	Structure ($)	Contractor ($)	Woodchips ($)	Supplies ($)	Total Costs ($)	Cost per Treatment Area ($ total ha^{-1} drained)
Greene Co.	$2,750.00	$5,250.00	$1,245.00	$500.00	$9,745.00	$512.35
Hamilton Co.	$1,640.00	NA[a]	$2,400.00	$350.00	$4,390.00	$216.96
Iowa 1	$1,970.00	$1,800.00	$3,350.00	$560.00	$7,680.00	$316.30
Iowa 2	$1,270.00	$1,890.00	$3,000.00	$780.00	$6,940.00	$428.73
Iowa 3	$1,640.00	$5,030.00	$4,650.00	$500.00	$11,820.00	$194.72
Iowa 4	$1,480.00	$2,710.00	$2,520.00	$400.00	$7,110.00	$585.64

[a] Contractor time donated.

from 12 to 60 ha, with the most expensive site on an area basis having the smallest drainage treatment area. The most expensive installation component for these bioreactors was either the contractor labor costs or the woodchips and transport, depending on the site. Ample local availability of woodchips can help minimize transport cost. Contracting fees from those who charge by the hour may eventually be reduced as increased experience with these systems may result in decreased installation time. Moreover, the cost of control structure manufacturing may decrease if there is a higher demand for these structures.

Using the average total influent load (24.2 kg N ha^{-1}) and the minimum or maximum percent load reduction (11.5% or 56.6%) from the NERF, Greene Co., and Hamilton Co. sites combined with the drainage treatment area and installation cost for each reactor allowed an estimation of cost efficiency. This simple cost evaluation, which assumed that the annual minimum or maximum percent load removals were maintained for 15 years, resulted in cost efficiencies of \$2.50 to \$12.30 kg N^{-1} and \$1.06 to \$5.21 kg N^{-1} for the Greene Co. and Hamilton Co. bioreactors, respectively. This range of values overlapped the denitrification bed cost efficiencies developed by Schipper et al. (2010a) of \$2.39 to \$15.17 kg N^{-1}. The lower end of values calculated here was also similar to cost efficiencies of other agricultural drainage water quality practices with reports of approximately \$2 to \$4 kg N^{-1} for wetlands and controlled drainage (Baker, 2009; Hyberg, 2007; Iovanna et al., 2008). Like these other practices, cost sharing for bioreactor installations in Iowa is available through the Environmental Quality Incentive Program with a one-time payment of \$3,999.50 (Iowa NRCS, 2010); this cost-share represents 34% to 91% of the total installation cost of the bioreactors in table 6.

CONCLUSIONS

Statistical analysis of the NO_3^- removal performance of four denitrification bioreactors treating agricultural drainage in Iowa showed that temperature and influent NO_3^- concentration were the most important factors for percent bioreactor N load reduction and N removal rate, respectively. Retention time was also a significant factor in percent load reduction for three of the four bioreactors, indicating that increased retention and warmer temperatures improve bioreactor NO_3^- removal performance. The inherently variable flow rates and drainage water temperatures over the course of a drainage season present a major bioreactor design challenge. Apart from design, however, active management may be able help optimize performance. For example, under cooler and higher spring flow conditions, retention times can be increased via control structure manipulation.

Overall, this work enhances understanding of denitrification bioreactors treating agricultural drainage by providing the first comprehensive performance evaluation of several such bioreactors in the U.S. Midwest. Averaged over all years, the four bioreactor sites had a mean removal rate of 2.3 g N m^{-3} d^{-1} and had bioreactor load and total load reductions of 45% and 32%, respectively. However, this mean removal rate may be low compared to other enhanced-denitrification systems, as some of the sites may have been nitrate-limited at times; removal rates may be greater for sites with higher nitrate influent concentrations or when non-ideal flow regimes are avoided (e.g., suspected short-circuiting at the NERF bioreactor). The simple economic assessment showed that at \$1.06 to \$12.30 kg N^{-1}, bioreactors had cost efficiencies comparable to other water quality technologies. More studies of field-scale performance from denitrification bioreactors designed with various methods and in various parts of the Midwest are needed to further improve understanding of the potential for these systems to positively impact water quality.

ACKNOWLEDGEMENTS

Acknowledgement for involvement with the NERF site is due to Ralph White and Ken Pecinovsky for sampling and bioreactor management and to Chad Ingels and the Coldwater Palmer Watershed Improvement Association for funding of the installation and monitoring equipment. Activities at this site were also funded by the Leopold Center for Sustainable Agriculture. The authors owe a debt of gratitude to Carl Pederson for field assistance with the NERF and Pekin sites and to Loren Shiers (retired) in the ISU ABE WQRL for sample analysis at these sites. Thanks to the Pekin-Packwood FFA chapter and Greg Brenneman for site management at the Pekin research site. Funding for the Greene County and Hamilton County bioreactor installation and monitoring was provided by Agriculture's Clean Water Alliance and the Sand County Foundation. Thanks to Reid Christianson for aid with flow analysis from all sites. This project was additionally supported by Agriculture and Food Research Initiative Competitive Grant No. 2011-67011-30648 from the USDA National Institute of Food and Agriculture.

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